**PRODUCT PERFORMANCE REVIEW**
**BY**
**KEVIN J. SWEENEY. SENIOR ENTOMOLOGIST, IB**

10/8/2009

Date: October 8, 2009

PM: Richard Gebken, PM 13

EPA Reg. No. 74843-2

Product Name: Buzz-Off Insect Shield Insect Repellent Apparel

Action Code: 570 – conditional data – non-PRIA

Dec #: 383199

DP: 344446

Active Ingredient: 0.52% permethrin

Formulation: RTU permethrin treated clothing

Site: Garments worn by humans

Pests: biting arthropods including mosquitoes and ticks for this action

PM request: review data submitted as a condition of registration to support increasing the number of washings from 25 to 70 on the label. Garments cannot be retreated or dry-cleaned.

Guideline: non-guideline OPPTS 810 series study

GLP studies? The studies are not GLP.

Data citation and submission:

For the original submission, under the selective method the registrant cited **MRIDs 46890401 and 46890402. The former was a justification for the use of in-vitro methods, while the latter was product specific analytical chemistry and in-vitro bioassay data collected without human subjects.** The other MRIDs submitted with the original submission (MRIDs 46890401 through 03) were unpublished reports from 1986-1987 originating with the USDA-ARS Insects Affecting Man and Animals Research Laboratory in Gainesville, Florida USA. These (latter) MRIDs were supplemental in nature because they did not contain product specific data. Instead, these studies discuss

how the in-vitro bioassay method was developed and validated. Much (but not all) of this data can also be found in other published and unpublished reports previously reviewed by EPA and the National Research Council.

The original submission included replicated bioassays with *Aedes aegypti* mosquitoes against permethrin treated end-use garments. These assays showed that >80% KD was achieved through 70 washings following a 2 minute exposure in a no-choice Petri dish assay. As a condition of this registrant, the registrant submitted a study with bioassays data against Anopheles, Ochlerotatus, and Culex mosquitoes and against two species of ticks. The fabric tested in these assays was the same as tested against *Ae. aegypti* in the original submission.

## Submitted study review:

**MRID 47210601 Buzz-off Insect Shield Insect Repellent Apparel: Efficacy against Three Species of Mosquitoes and Two Species of Ticks by Benzon Research, Inc. Completed on July 12, 2007.**

Purpose: To determine the knockdown of mosquitoes and ticks with the subject product using an in-vitro assay to satisfy a condition of registration.

Materials and Methods:

Tested mosquito species: Adult life stage of *Culex quinquefasciatus, Anopheles quadrimaculatus, and Ochlerotatus taenioryhnchus* tested through 70 washes. *Aedes aegypti* tested through 25 washes to add a data point to the original data with the same materials.

Tested tick species: the blacklegged tick, *Ixodes scapularis*, and the brown dog tick, *Rhipecephalus sanguineaus.*

Procedure: The procedure was the same one as used in the previous study. The Petri dish lid was laid on the lab table surface. A fabric swatch was laid inside and over the lid. A hole was made in the bottom of the Petri dish. The bottom of Petri dish was laid on top of the fabric to obtain a snug fit. Some of the fabric swatch hung over the side. For mosquitoes (10-20) were aspirated into the dish. A piece of tape was placed over the hole. For ticks, 10-15 chilled ticks were transferred to the dish. The procedure was repeated for treated and untreated swatches. Mosquitoes or ticks were exposed for two minute. After this time the plate was held vertically, tapped on the fabric side, and the swatch was slowly removed. Any remaining threads were removed with fine forceps. The number of injured mosquitoes was recorded. This number was subtracted from the total in the plate.

Mosquitoes or ticks remained in the dishes and knockdown was recorded at 15 and 60 minutes post-exposure. Plates were inverted at each counting period. If a mosquito or tick could not right itself after plate inversion - or was moribund or not moving - it was

counted as knocked down. After the knockdown counts were completed, mosquitoes or ticks were frozen and the total in the dish counted and recorded.

**Treatments:** Buzz-off Insect Shield Repellent Apparel (EPA Reg. No. 74843-2) (BOIS Process using BOC-14D was used) Samples were 50:50 nylon/cotton Army Combat Uniform Trousers from Lots 23 and 31. Samples from the same lots were used in the previous study with *Aedes aegypti*, which was accepted in 2006. Ten samples of product fabric were tested at each of the wash durations for each species as explained below. A large sample of residue chemistry data were submitted, reviewed and accepted from MRID46890402 to verify the existence of permethrin in the fabric samples.

Mosquitoes:

*Aedes aegypti* – 12 samples from Lot 23 were tested through 25 washes to supplement the data reported through 70 washes in MRID46890402. Each replicate consisted of 13-27 mosquitoes. One untreated control was tested.

*Culex quinquefasciatus* – 10 samples from Lot 23 through 25 washes; 8 samples from Lot 23 through 50 washes; 1 sample each from Lots 31 and 32 through 50 washes; and 9 samples from Lot 23 through 70 washes. Each replicate consisted of 9-18 mosquitoes. One untreated control was tested.

*Anopheles quadrimaculatus* – 10 samples from Lot 23 through 25 washes; 8 samples from Lot 23 through 50 washes; 1 sample each from Lots 31 and 32 through 50 washes; and 10 samples from Lot 23 through 70 washes. One untreated control was tested. Each replicate consisted of 9-17 mosquitoes. One untreated control was tested.

*Ochlerotatus taenioryhnchus:* 10 samples from Lot 23 through 25 washes; 8 samples through 50 washes; 1 sample each from Lots 31 and 32 through 50 washes; and 10 samples from Lot 23 through 70 washes. Each replicate consisted of 14-20 mosquitoes. Two untreated controls were tested.

Ticks:

*Ixodes scapularis* – 10 samples from Lot 23 through 25 washes; 1 sample each from Lots 31 and 32; 8 samples from Lot 23 through 50 washes; and 10 samples from Lot 23 through 70 washes. The number of ticks per replicate equaled 12. One untreated control was tested.

*Rhipecephalus sanguineaus* - 10 samples from Lot 23 through 25 washes; 1 sample each from Lots 31 and 32 through 50 washes; 8 samples through Lot 23 through 50 washes; and 10 samples from Lot 23 through 70 washes. Each replicate consisted of 12 ticks. One untreated control replicate was tested.

**Results:**

The results were reported at 15 and 60 minutes post-treatment exposures in terms of the number of mosquitoes or ticks knocked down and as % knockdown for each replicate. Results were summarized in table form to report the mean 60 minutes knockdown values. (The % knockdown value of regulatory concern when evaluating the success of the test is ≥ 80% knockdown.) Mean KD values for mosquitoes and ticks exceeded 90% for all washes and species except for *Aedes aegypti*, where the 70 wash value achieved 80% KD. 100% of *Ixodes scapularis* ticks were knocked down by 60 minutes post-exposure. Fifteen minute knockdown was very good for most species. Brown dog ticks were not knocked down as quickly, probably owing to their large size.

**Conclusion:** These data show that the subject product causes knockdown of tick and mosquito species following a two minute exposure to the treated fabric. Based on these data the permethrin in the fabric was bio-available to provide protection from target pests.

From my last review:
"Based on already published data, permethrin treated fabric is effective at levels against pests as low as 0.01mg/cm$^2$. These data do not contradict this finding but appear to show that even higher levels of permethrin in fabric may not be available to KD mosquitoes. The subject submission provided bioassay data with mosquitoes in a no-choice test and also reported GC data collected on lots of Buzz-Off treated fabric, including swatch samples and whole garment analyses. These data show no clear trend between the residue data submitted and bioassay success. However, the bioassay data show that the more washes performed, the less successful the treated garment is in knocking down mosquitoes. This result is consistent and less variable than the residue data, which indicates a fairly high level of permethrin in fabric even through one hundred washes. The levels vary between washes.

The bioassay and analytical chemistry must be examined together. The bioassay has been shown to be the more sensitive indicator of product success in this study and supports the registrant claim that the presence of permethrin alone may not indicate that it is available to repel/KD the target pest. However, the EPA should always require registrants to show permethrin residue present in treated garments and never rely on bioassay data alone."

**Recommendations:**

1. I recommended that the registrant change: "Repels mosquitoes (including those that transmit West Nile virus)" to "Repels mosquitoes". As stated in my 2006 review this claim can be revisited after submission of conditional efficacy data. The efficacy data now support this claim.

2. These data satisfy the conditions set forth in 2006. However, this study may not completely satisfy the PDCI issued for permethrin treated fabrics because: 1) different fabric types may affect repellency and bite protection; and 2) repellent studies for

permethrin have used % repellency as a standard of efficacy instead of complete protection time. Permethrin treated clothing is making many of the same label claims found on skin-applied products. The standard of success for skin-applied products is Complete Protection Time and confirmed landings, not bites, are used as the endpoint.